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**Research Article**

# BACTERIOLOGICAL PROFILE OF URINARY TRACT INFECTIONS DURING PREGNANCY IN GAROUA, NORTHERN CAMEROON REGION

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**ABSTRACT**

Urinary infection during pregnancy is a common condition and can have significant complications on the mother and the child. General objective: The aim of our study was to determine the profile of the bacteria isolated and assess their sensitivity to antibiotics in urinary infections in pregnant women. We conducted a descriptive and cross-sectional study for four months on the urine culture on 160 pregnant women whose samples were analyzed at the Laboratory of Bacteriology Centre Pasteur of Garoua. The identification of bacteria was carried out using conventional bacteriological tests Antimicrobial susceptibility according to the disk diffusion method has been achieved, as recommended by CA-SFM 2015. Urinary tract infection was found in 21.87% of cases. The study of the patients' age and parity enabled noted predominance among paucipares 40.00%, while all ages can be affected, predominantly between 20 and 29 years. The study of the occurrence of dates showed a frequency in the 2nd trimester of pregnancy. The predominant species were *Entérobactériaceaes* 60% *Escherichia coli, Klebsiella pneumoniae* 14.3% and Gram-positive *cocci* with 20% of cases. Rates *Entérobactériaceaes* sensitivities were 81.6% respectively, for the amoxicillin clavulanic acid, 85.7% to 85.7% and cephalothin 3GC (cefotaxim, cetriazon). Susceptibility to imipenem and nitrofuran were 85.7% and 100% respectively. However, the presence of producing *Enterobacteria bêta-lactamases* broader spectrum including *Escherichia coli,* must be taken into account in our local epidemiology for the development of therapeutic algorithms. The molecular characterization of these strains is needed to study the different enzymes involved in resistance to beta-lactams.The frequency of urinary infections during pregnancies requires a regular and systematic tracking all pregnant women. This will make it possible to accompany and as well as possible guide the assumption of responsibility of the producing stocks of *beta-lactamases* with widened spectrum and probably to reduce the mechanism of resistance to antibiotics.



## INTRODUCTION

Urinary tract infection (UTI) is the bacterial colonization of the urine and / or urinary tract from the kidneys to the urethral meatus (Fourmi and *al*,1996). It is the second most common infectious disease contracted by humans after respiratory diseases. They are found in children, adults and the elderly, in both sexes (Abalikumwe,2004 ; Letonturier,2006). It remains one of the most common complications of pregnancy and its prevalence varies from country to country depending on the strata of the population concerned (Mergerand *al*,1995). It is estimated that more than 8 million cases of UTI are reported each year and more than 1 million cases of hospitalization at an annual cost of more than $ 1 billion (Matuszkiewicz and *al,*2015). These infections became common during pregnancy due to age (one in three women of childbearing age has a urinary tract infection) and physiological changes related to pregnancy (Lansac and *al,*2003) This could be explained more by the physiological, mechanical and hormonal characteristics of pregnancy favoring urinary stasis. To this specific phenomenon of pregnancy could be added contamination of the short urethra by gastrointestinal and perineal bacteria (Kladensky,2012). It is a real public health problem that can lead to both fetal, neonatal and maternal risks such as premature birth, fetal mortality, anemia, pyelonephritis and chronic renal failure; without forgetting the therapeutic complications that can generate the phenomena of resistance of

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germs to antibiotics (Vasudevan,2014 ;Masinde and *al,*2009). Despite the often multifactorial origin, each pregnant woman has a very specific reason for developing cystitis and one must look for the triggering factor or factors of bacterial origin (Sheffield and *al*,2005).Gram negative bacteria are the main cause of these urinary tract infections (Battikhi and al,2015). Among them, *Enterobacteria* comes first with Escherichia coli (85% infections outside the hospital), followed by *Klebsiella spp*, *Proteus spp, and Pseudomonas spp* (5-10%); *Staphylococcus saprophyticus* (5-15%) for Gram positive (Emiru and *al* 2013 ; Unlu and *al,*2014). Most of these uropathogens were identical over the years, but the pattern of antibiotic resistance was varied (Van Hoek and *al,*2011). Although urinary tract infection has a clinical translation, its asymptomatic form remains frequent during pregnancy which can progress to rough forms and be the basis of all possible complications and more particularly by multidrug-resistant microbes (Lee and *al*,2008 ;Tadesse and *al,* 2014) . The diagnosis of UTI constitutes a major workload in medical analysis laboratories, where *Escherichia coli* reveal the highest predominant rate although, other reports have shown that the causal organism of UTI changes during the year and other microorganisms wereresponsible for infection (Emilie and *al*,2011). The management of these infections in terms of screening and treatment still poses some problems. We can note the prescription for urine pellet examination, therefore without bacterial identification or antibiogram. This is partly due to the high cost of investigating and treating UTIs for most pregnant women. The isolation of pathogens associated with UTI and the determination of their antibiotic-sensitive pattern will potentially reduce the inappropriate prescribing of antibiotics and resistance (Foxman and *al*,2002). The fight against UTI during pregnancy requires health education and systematic screening (clinical and biological) of this condition in all pregnant women. Cytobacteriological urine examination (CBUE) with the antibiogram is the best biological test for the diagnosis of urinary tract infection. Appropriate treatment based on the results of the antibiogram is the rule. To do this, what is the profile of germs from UTI during pregnancy in our region? The aim of the study is to define the sensitivity to antibiotics of microbial strains isolated from pregnant women with infection of the urinary tract.

## METHOD

This is a descriptive and cross-sectional study on CBUE in 160 pregnant women, whose samples were analyzed at the Bacteriology laboratory of the Center Pasteur du Cameroun Annex in Garoua. The study was carried out over a period of four months, from 05 May to 05 September 2015. It involved all pregnant women who attended the Garoua Regional Hospital during prenatal consultations during the period of our study. Were included in this study, pregnant women coming in prenatal consultation to Garoua Regional Hospital during the period of our study regardless of the age of pregnancy and having given their consent. Were excluded in our series, any pregnant woman who consulted outside this period and who did not consent to participate in the study after an interview. This is a non-probability convenience sampling, simple random type.

***Bacteriological study***

The sample was taken in a sterile bottle with stopper. Each urine sample was subjected to a routine cytobacteriological examination comprising

A direct examination to assess Leukocyturia and the figured elements of urine (red blood cells, crystals, cylinders ...) using an optical microscope and counting cells of the KOVACS

Uroculture with count of germs (Bacteriuria). Thus, for the realization, different materials and media were used: sterile bottle (for sampling); box ofkneaded; Volumetric balloon; alcohol lamp; incubator; sterile swab; pliers; jar and sterilizer (Paupinel, Autoclave) and reagents for gram staining (gentian violet, lugol, alcohol-acid, and carbolic red); kits for grouping streptococcus (Streptoplus) and for staphylococcus aureus (Slides staph); culture media of the CLED; Sabouraud agar for the isolation of yeasts and Muller Hinton agar for carrying out antibiograms. The samples were incubated at 37° C for 18-24 h. The presumptive identification was made by cultural morphology, and supplemented by biochemical characters (galleries Api 20 E, Slidex Staph, Slidex Strepto). ***UTI's*** *biological diagnosis was based on Kass criteria. Leukocyturia ≥104 / ml + bacteriuria ≥105ufc / ml.*

***Antibiotic sensitivity study***

A standard antibiogram was carried out using the diffusion technique in agar medium. The disc inhibition diameters were measured using a caliper and the interpretation was made according to the standards of the antibiogram committee of the French Microbiology Society (CA-SFM 2014) (JC Soussy 2014). The antibiotics usually used to treat infections in pregnant women have been selected; 12 antibiotics were tested: Ampicillin (10ug), Amox + clav acid (20 / 10µg), Cefotaxim (30µg), Ceftriazon (30µg), Cefoxitin (30µg), Imipénèm(10µg), Fosfomycin (50µg), Nitrofuran (300µg), Erythromycin (15UI), Pristinamycin (15µg), Oxacillin (5µg), Rifampicin (30µg), Ticarcillin (75 µg). When a synergistic image appeared, the Synergism Test was used to better illustrate the detection of ESBL, by placing around the disc of amoxicillin + clavulanic acid, the discs of Cefepim, Cefotaxim, and Ceftazidim , at a distance of 3cm from center to center, according to the recommendations of the French Antibiogram Committee of the French Microbiology Society (CA-SFM 2015).

**Table I:** Distribution of women with urinary tract infection according to socio-demographic characteristics

| Characteristics | Staff | Frequencies (%) | P-Value |
|---|---|---|---|
| **Old** | | | **P = 0,02717** |
| = 19 | 01 | 2,85 % | |
| 20-29 | 28 | 80 % | |
| 30-39 | 04 | 11,42% | |
| = 40 | 02 | 5,71% | |
| **Profession** | | | **P = 5.609e-07** |
| Housewife | 23 | 65,71 % | |
| Student | 06 | 17,14 % | |
| Civil servant | 04 | 11,43 % | |
| Others | 02 | 5,71 % | |
| **Marital Statut** | | | **P = 0.0001** |
| Married | 29 | 82,86 % | |
| Single | 06 | 17,14 % | |
| **Education level** | | | **P = 0,0141** |
| Out of school | 15 | 42,86 % | |
| Primary | 11 | 31,43 % | |
| Secondary | 07 | 20 % | |
| Higher | 02 | 5,71% | |
| **Pregnancy age** | | | **P= 0,0002** |
| 1st Trimester | 05 | 14,19 % | |
| 2nd Trimester | 23 | 65,71% | |
| 3rd Trimester | 07 | 20 % | |
| **Parity** | | | **P= 0,0490** |
| Nulliparous | 11 | 32,43 % | |
| Primiparous | 07 | 20 % | |
| Pauciparous | 14 | 40 % | |
| Multiparous | 03 | 8,57 % | |

**Table II:** Distribution of pregnant women with UTI according to the symptomatology

| Symptoms | staff | Frequencies (%) |
|---|---|---|
| Dysuria | 23 | 65,71 % |
| Voidind burn | 19 | 54,28 % |
| Pollakiuria | 16 | 45,71 % |
| Polyuria | 05 | 14,28 % |
| Pelvic pain | 15 | 42,85 % |
| Low back pain | 13 | 37,71 % |
| Fever | 07 | 20 % |

**Table III:** Distribution of isolated species in the UI during the study period

| Species | staff | Frequencies(%) |
|---|---|---|
| BG(-) 80% | | |
| *E.Coli* | 21 | (60 %) |
| *K.Pneumoniae* | 05 | (14,3 %) |
| *P.Mirabilis* | 01 | (2,9 %) |
| *C.frundii* | 01 | (2,9%) |
| CG(+) 20% | | |
| *S.Aureus* | 03 | (8,6 %) |
| *SCN* | 02 | (5,7 %) |
| *Streptococcus b* | 02 | (5,7 %) |
| | 35 | 100 % |

**Table IV: Overall antibiotic sensitivity profile of all isolated germs**

| ANTIBIOTICS | Number (%) of susceptible strains | | | | | | |
|---|---|---|---|---|---|---|---|
| | *E.coli* | *K.Pneumonia* | *P.Mirabilis* | *C.frundii* | *Staph Aureus* | *SCN* | *Strepto b* |
| Ampicillin | 0(0) | 5(100) | 1(100) | 1(100) | 2(66,7) | 1(50) | 2(100) |
| Amo /Ac Clav | 16(76,2) | 5(100) | 1(100) | 1(100) | 3(100) | 2(100) | // |
| Oxacillin | // | // | // | // | 3(100) | 2(100) | // |
| Cefalotin | 17(81) | 5(100) | 1(100) | 1(100) | // | // | 2(100) |
| Cefotaxim | 18(85,7) | 5(100) | 1(100) | 1(100) | 3(100) | 2(100) | 1(50) |
| Ceftazidin | 17(81) | 5(100) | 1(100) | 1(100) | // | // | // |
| Ceftriaxon | 18(85,7) | 5(100) | 1(100) | 1(100) | 3(100) | 2(100) | // |
| Cefoxitine | // | // | // | // | 3(100) | 1(50) | // |
| Erythromycin | // | // | // | // | 3(100) | 2(100) | 1(50) |
| Nitofuran | 21(100) | 5(100) | 1(100) | 1(100) | // | // | // |
| Lincomycin | // | // | // | // | // | // | 2(100) |
| Imipénèm | 21(100) | 5(100) | 1(100) | 1(100) | // | // | // |
| Rifampicin | // | // | // | // | 3(100) | 2(100) | 1(50) |
| Fosfomycin | // | // | // | // | 3(100) | 2(100) | // |
| Pristinamycin | // | // | // | // | 3(100) | 2(100) | // |
| Ticarcillin | 0(0) | 0(0) | 0(0) | 0(0) | // | // | // |

**Table V: Distribution of ESBL producing strains.**

| BACILLE GRAM (-) | Staff | ESBL(-) N/(%) | ESBL(+) N/(%) |
|---|---|---|---|
| *ENTEROBACTERIACEAES* | 28 | 25 (89,28%) | 03 (10,71%) |
| *Escherichia coli* | 21 | 18 (85,71%) | 03 (14,28%) |
| Others *Entérobactériaceae* | 07 | 07 (100%) | 0 (0%) |

***Quality control on sensitivity tests***

This control was carried out with the so-called "reference strains". Whenever there was a new set of reagents, media, or device tuning, these strains were used: *Escherichia coli* ATTC 25922; *Klebsiella pneumoniae* ATCC 700603; *Staphylococcus aureus* ATCC 25923; *Streptococcus group b* ATCC 12386.

The data collected was encoded on a spreadsheet in Excel software (Microsoft, USA, 2010) and then imported for processing on R.2.13 computer software. These data are presented and described according to qualitative variables (numbers, percentages and 95% confidence intervals). Comparative analyzes were made using Test Chi 2. The significance threshold used was 5%. A research certificate had been obtained before the start of our study. We also obtained authorization from the various managers of the structures concerned to conduct our research. This study was carried out with informed consent without financial contribution from patients, in strict compliance with medical confidentiality.

## RESULTS

In total, we took 160 urine samples from pregnant women for cytobacteriological urine examinations, of which 35 were carriers of bacterial urinary tract infection, a frequency of 21.87%. The 20-29 age group was the most represented with 80% of the cases. The extreme ages were 19 and 45 with an average age of 25.8 ± 5.61 years. Pregnant women not in school were in the majority with 42.86% of cases. Married women in our study had a frequency of 82.86%. The urinary tract infection was the preserve of the eyelids (40.00%), occurring in the second trimester of pregnancy (65.71%). The most recurrent symptoms were represented by these triads: Dysuria 60%, voiding burn 57.14% and pollakiuria 48.57%. *Enterobacteriaceae* were frequently encountered, with *Escherichia coli* 60% of cases, followed by *Klebsiella pneumoniae* 14.3% cases. *Staphylococcus aureus* is the most common in Gram-positive cocci with 8.6% of cases. 03 isolates were BMR. All were ESBL-producing E. coli and represented approximately 10%.

*Escherichia coli* showed good sensitivity to Nitrofuran, and Imipenèm 100% to cephalosporins. There is also a significant sensitivity to AMC 76.2%. The resistance to Ampicillin and Ticarcillin was 100%. All strains of *Klebsiella pneumoniae* were susceptible to cephalosporins AMC, Nitrofuran, Imipenem 100%. All strains of *staphylococcus* showed good sensitivity to all of the antibiotics tested. However, 33.3% resistance to Ampicillin was noted for *Staphylococcus aureus* and 50% for *Staphylococcus* with Coagulase Negative (SCN). Group *b Streptococcus* had a moderate sensitivity to Erytromycin, Rifampicin with a rate around 50% each. The highest sensitivity was observed for Ampicillin and Cefalotin with a rate of 100% except for Lincomycin.

## DISCUSSION

During our study, we recorded 35 cases of bacterial UTI on a sample of 160 urines from pregnant women, a frequency of 21.87%. Our results are similar to those of other studies (Mokube and *al*,2013;Stamm and *al*,2001) which found a frequency of 23.5% and 23% respectively of cases of pregnant urinary tract infection. But other studies report a rate of 12.7% (Alassane, 2012). It emerged in our study that the most affected subjects were between 20-29 years of age, with an average of 25.8 years and represented 80% of the whole. Statistical analysis has shown that age has an influence on the occurrence of pregnancy-positive urinary tract infection. This observation was also made by an author (D'Ercolle and *al*,1993), where he found that 78.72% of infected parturients had an age that varied between 20-29 years. As for Mokube and*al*, he declared that 68% of pregnant women were between 21-30 years old. The influence of age can be explained by the fact that this period is the one in which the woman exhibits active sexual activity. Brides were the most represented with 82.86%. This rate is similar to that of Mokube and *al*, who found 66% of pregnant women who were married. This can be explained by the fact that married women are pushed by their husbands, who give them the means for these prenatal visits. Urinary tract infection was much more observed in pregnant women with less

education (42.6%). Indeed, the female gender in our country in general and in our region in particular is undereducated. Under-schooling is a handicap for good hygiene and for hygiene control in pregnant women who are prone to much more infection. We found that there was a significant difference between the occurrence of UTI in pregnant women and their level of education; thus, there were 31.43% of cases among those who had primary education, 20% of cases among those who completed secondary education and 5.71% among those who completed tertiary education. So therefore, we see that urinary tract infection seems to decrease with the level of education in pregnant women. In our study, 65.71% of urinary tract infections are observed in the second trimester of pregnancy. Indeed, urinary tract infection is observed much more in pregnant women from the second trimester of pregnancy, because progesterone, present in large quantities, decreases the tone and contractility of the smooth muscle fibers of the ureter. It inhibits ureteral peristalsis and thus promotes stagnation of urine and vesicoureteral reflux. It decreases the sphincter tone, promoting urethro-vesical reflux. Estrogens are responsible for hyperthermia of the trigone. They also promote the adhesion of germs to the urothelium (Njeh and *al*,1996 ; Haider and *al*,2010). Physico-chemical factors lead to increased concentration of sugars and amino acids, alkalization of urine.These modifications make the environment more favorable to the development of germs. They become more marked from the second trimester, hence the appearance of symptoms of urinary tract infection from the second trimester of pregnancy (Haider and *al,* 2010 ;Paban and *al,*2007). The largest number of women come for consultations from the second trimester of pregnancy and it is during these antenatal consultations (ANC) that they are asked for a follow-up report on pregnancy which is the CBUE. And as the urinary tract infection of pregnant women sometimes is asymptomatic, this is why we discover cases of urinary tract infections without frank manifestation. Other authors (Stamm and *al,*2001; Layla,2003 ;Karhate,2011) rather noted a high rate of UTI in the third trimester of pregnancy with respectively 63.7%, 79% and 78.40% of the cases. Indeed, we note that women come late for antenatal consultation either for lack of means or through ignorance; which explains a higher number of women seen in the second trimester or third trimester, and consequently a higher number of cases of urinary tract infections. We observed a high rate of urinary tract infection in pauciparous people (40%). This same observation was observed by certain authors (Alassane, 2012 ; Demba,2007).] who reported a rate of 46% and 47.2% respectively in Mali. This observation seems to vary from one author to another, all the more so than Koné. H (Koné,2002), observed a high rate of 40% rather in primiparous women, while Mokube and *al* concluded an increase in frequency in multiparous women having much more close pregnancies with a low level of hygiene. All this leads us to believe that the infection can be observed in any woman. The clinical signs were made up of usual functional signs of a urinary tract infection: dysuria 60%, micturition burn 57.14%, and pollakiuria 48.57%. Our results corroborate with those of the literature (Alassane, 2012 ;Traore and *al*,1990). The most represented germ was *Escherichia coli* with 60% followed by *Klebsiella Pneumonia* 14.3%. For Mc Gready, *Escherichia coli* was 87.5% of the cases and *Klebsiella Pneumonia* 3.12% of the cases (McGready and*al,* 2010). Layla Benomar reported a rate of 53.7%, for *Escherichia coli*, followed by *Klebsiella Pneumonia* with a rate of 13.40% of cases. In the Gram-positive cocci group, the isolated germs were: *Staphylococcus aureus* 8.6%, SCN *and Streptococcus b* with 5.7% each. pregnancy does not confer any bacterial specificity, the germs found during pregnancy are in the same proportions of urinary tract infections in general. *Enterobacteriaceae* have shown sensitivity to beta-lactams, the rate of which varies according to the bacterial species and the molecule of antibiotic. The 1st and 3rd generation cephalosporins have generally shown good activity on *Enterobacteriaceae* species. This observation is similar to that of the authors (Lamia and *al,*2010). But we have already observed the appearance of resistances on these molecules. This decrease in sensitivity could find a justification in the self-medication and anarchic manipulation of thesemolecules in our society. Among the antibiotics tested, the most active on *Escherichia coli*in our study were: 3GC (Cefotaxim, Ceftazidim, Ceftriazon,), 1GC (Cefalotin), Nitrofuran, Imipenem and AMC. Arzouni and Tahirou had the same antibiotics active on *Escherichia coli* (Tahirou ,2005). Among the E. coli isolated during our study, we found 3 (14.28%) ESBL-producing strains; which shows the circulation of multi-resistant strains in our regions, despite the small sampling. Already in the early 2000s, the resistance of *Escherichia coli*isolated from urinary 3GC infections (cefotaxim or ceftriaxon) was low: it was less than 1% in 2003 and 4% in 2008 according to AFORCOPI-BIO (De Mouy and *al* ;2007a,2008b). In our study, we noted a strong emergence of ESBL producing strains. In order to limit this spread, it would be wise to conduct a justified antibiotic therapy policy and / or a restriction in the prescription of third generation cephalosporins and even all cephalosporins, to lead to a significant decrease in the frequency of ESBL. Such directives are not yet in force in Cameroon. In the current state of the acquisition and distribution of antibiotics in our regions, it is unlikely that we will soon have new effective molecules on ESBL-producing *Escherichia coli*and other multi-resistant *Enterobacteriaceae*. The fight against the emergence of ESBL producing *Escherichia coli* is now not only a problem but also a public health duty. The sensitivity of *Staphylococcus aureus* was 100% for most antibiotics except for Ampicillin where it was reduced to 66.7%. In contrast, Alassane Sangaré had a varied sensitivity to the antibiotics tested: 82.2% for AMC, 71% for Cefalotin, 98% for Prisnamycin, 50% for Ampicillin and Cefoxitin 50%. The *Coagulase negative staphylococcus* strains were sensitive to Ampicillin, AMC, Cefalotin, Oxacillin, Nitrofuran to Pristinamycin to Erytromycin 100%. *Streptococcus b* were sensitive to Ampicillin and Cefalotin 100% and 50% to Cefotaxim, erythromycin, and Rifampicin. However, 100% total resistance for Lincomycin has been noted. No explanation can be given, especially since the number is small (02), we can not conclude anything.

## CONCLUSION

The frequency of UTI during pregnancy is high (21.87%) in our region. This study also showed that the most bacterial isolates were *Enterobacteriaceae* among which *Escherichia coli*(60%). Therefore, a systematic diagnosis in all pregnant women is necessary in order to provide treatmentappropriate necessary. The CBEU accompanied by an antibiogram remains

the best examination for the diagnosis of urinary tract infections. In our series, 10.71% of urinary tract infections in pregnant women are BMR. Multicentric prospective studies should be carried out to confirm these worrying results.

**Authorship contribution**

All authors contributed to the designing, preparation, editing and fnal review of the manuscript

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**Confict of interest**

The author declares that there is no confict of interest

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